Pulsed Transdermal Electrical Stimulation (pTES):

An Overview

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Note: The Neuros™ pTES device is not yet currently available for sale in the EU market. The product will be made available in the EU subject to the receipt of the CE Mark.

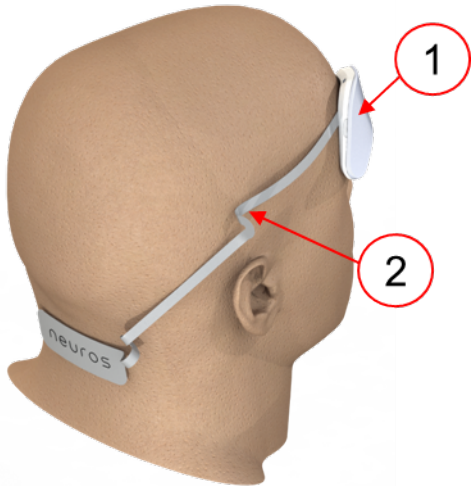
1.0 SUMMARY

The Neuros (Model 1.0) Pulsed Transdermal Electrical Stimulation (pTES) System is a Cranial Electrical Stimulator (CES) which is intended for the alleviation of insomnia and anxiety. Neuros transdermally applies a light bi-phasic electrical current to modulate the trigeminal and the cervical nerves to suppress sympathetic nervous system activity thereby reducing anxiety and improving sleep quality. The device has been marketed as a lifestyle product within the United States under the brand Thync and mentions of clinical data in this White Paper may reference the Thync name. The purpose of this White Paper is to introduce Neuros pTES technology along with an analysis of the existing literature surrounding (CES) devices.

CES devices have a long history of clinical research and use. In general, these devices are effective at alleviating symptoms of anxiety and insomnia. There is evidence to suggest CES is also effective for the treatment of depression and pain, however neither of these claims are made for Neuros.

This White Paper will present actual clinical data collected with Neuros which demonstrate that it is safe, minimizes side effects of discomfort, and is effective at reducing symptoms of anxiety and insomnia. Additionally, nearly 50 years of clinical research data has been fully appraised and evaluated for safety and performance. These data are further synthesized to provide objective and measurable evidence of safe, effective therapy for the treatment of anxiety and insomnia.

2.0 THE DEVICE



2.1.1 Tradename and Model: Neuros Model 1.0 (note: a precursor version of this device has been marketed in the USA and Canada under the brand name "Thync". Neuros brand will be made available outside of North America)

2.1.2 Device Type: Pulsed Transdermal Electrical Stimulation (pTES) System

2.1.3 Device Group Type: Cranial Electrical Stimulator (CES)

2.1.4 Device Overview

The device consists of two principal hardware parts as shown; the durable Module (1) which contains the driving electronics, rechargeable battery (source of power), and connections with the limited-use Strip Electrode Assembly (2) (also referred to as the Strip). The strip contains the conductive electrodes at the trigeminal nerves (V1, V2) at the forehead and the cervical nerves near vertebrae C2 and C3 at the back of the neck. The strip also contains a secondary adhesive for holding the device onto the skin of the wearer. The strip can be used for at least one therapy but can be re-used as necessary until the adhesive is no longer able to support the weight of the device.

The device uses low-energy, pulsed electrical currents (up to 4 mA) to favorably regulate the sympathetic and parasympathetic nervous systems for enhanced relaxation and sleep. With pulsed electronic energy at greater than 400 Hz applied to these afferent pathways, the parts of the brain which control physiological arousal, regulate stress and anxiety, and modulate sleep quality and quantity are stimulated to target various brain regions via 'bottom-up' pathways, including the cortex, limbic system, and thalamus. Activity of the sympathetic nervous system is inhibited, thereby decreasing stress and anxiety while improving sleep quality.

The device is controlled by a smartphone application which communicates with the Module via Bluetooth®. A treatment lasts 15 minutes and is best used by the patient prior to going to bed. Initial positive results are generally reported after repeated daily sessions ranging from 2-14 days, depending on the individual.

2.2 Theory of Operation

[This section has been adapted from the introduction in Boasso et al. 2016. [1]]

The device provides a method of modulating psychophysiological arousal and stress responses by providing electrical signaling waveforms through afferent pathways of cranial nerves to neuromodulatory nuclei in the brainstem. During signal transmission to cortex, incoming sensory signals carried by the trigeminal (V1 – V3)

nerves simultaneously undergo local processing by a series of highly interconnected structures including nuclei of the ascending reticular activating system (RAS) located in the pons.

Figure 9.5: Neuros Interface of the Ascending RAS through the TNSC

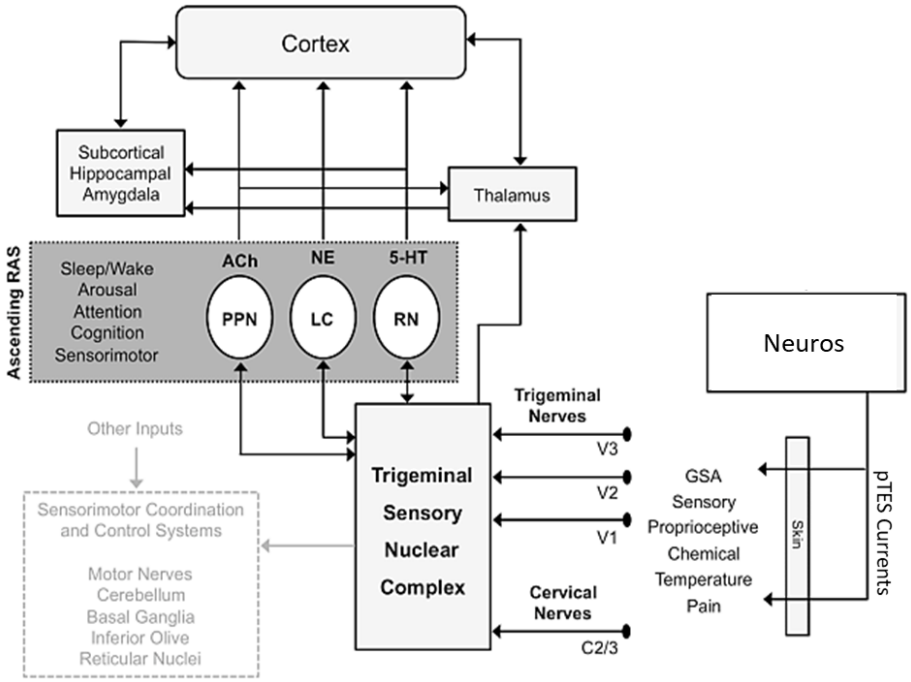


Figure adapted from Boasso, et al. 2016

The ascending reticular activating system (RAS) is a collection of nuclei and circuits that sort, filter, integrate, and transmit incoming sensory information from the brainstem to the cortex to regulate sleep/wake cycles, arousal/alertness, attention, and sensorimotor behaviors. [2, 3, 4, 5, 6, 7, 8, 9] The endogenous neuromodulatory actions of the RAS on consciousness and attention are orchestrated by at least three distinct sets of brainstem nuclei that include cholinergic neurons of the pedunculopontine nucleus (PPN), noradrenergic neurons of the locus coeruleus (LC), and serotonergic neurons of raphe nuclei (RN). [2] These pathways transmit neuromodulatory acetylcholine (ACh), norepinephrine (NE), and serotonin (5-HT) signals to higher-order brain structures to gate attention and regulate awareness, arousal, and sleep.

Through a cytoarchitectural meshwork of interconnected brain stem nuclei in the pons and midbrain, sensory inputs first act upon the brain to engage ascending RAS networks, which generate global arousal ("waking"), alerting, and orienting cues to parsed sensory information, which projects through thalamic pathways onto the cortex for additional processing and integration. More specifically, the local and distal synaptic circuits formed by axons of neuromodulatory RAS networks (including neurons of the LC, PPN, RN) gate information flow from the sensory environment to the cortex and, in an activity-dependent manner, are capable of rapidly triggering neurobehavioral transitions across different states of behavioral awareness and consciousness. [8, 10, 11, 12] Depending on their firing rates, neurons of the PPN can differentially mediate REM sleep states [2, 13] and neurons of the LC can trigger sleep/wake transitions. [14, 15] Disrupted activity of ascending RAS networks underlies several neuropsychiatric conditions and disorders, such as insomnia, anxiety, depression, posttraumatic stress disorder (PTSD), and attention deficit hyperactivity disorder (ADHD). [2, 16, 17, 18]

The trigeminal nerve or the fifth cranial nerve (cranial nerve V) bilaterally innervates the anterior half of the head and face including around the top of the scalp, the forehead, around eye orbits, nasal region, lips, jaw, and the oral cavity. Three main branches of the trigeminal nerve (ophthalmic (V1), maxillary (V2), mandibular branches (V3)) and their thousands of sub-branches transmit sensory information (chemical, thermal, mechanical, pain, and proprioceptive) via monosynaptic connections to the trigeminal sensory nuclear complex (TSNC). The TSNC itself is an elongated structure with several functional divisions (for example, the primary sensory nucleus and the spinal nucleus) spanning from the cervical spinal cord to the midbrain. The TSNC has been functionally mapped

using multimodal trigeminal stimulation combined with functional magnetic resonance imaging (fMRI) and diffusion tensor imaging (DTI) in humans. [19, 20]

The TSNC also receives some non-trigeminal sensory information from the neck via cervical afferents, which for the device are via the cervical (C2/C3) nerves. [21] In turn the TSNC projects incoming sensory information through ascending pathways to multiple brain regions that regulate arousal and coordinate neurobehavioral engagement with the environment, such as the thalamus , [22, 23] the superior colliculus, [22, 24, 25] the cerebellum, [24, 26] and the inferior olive. [24, 27] Several other electrophysiological and neuroanatomical studies have provided definitive evidence of functional synaptic connectivity between trigeminal afferents and the PPN and LC. [28, 29, 30, 31]

Besides its robust functional connectivity to ascending RAS networks, a major advantage of the TSNC as a neuromodulation target is that its primary monosynaptic inputs can be noninvasively accessed and coupled to using safe and comfortable transdermal neurostimulation approaches. Transcutaneous trigeminal nerve stimulation (TNS) has been shown to be effective for treating neuropsychiatric conditions like depression, [32] PTSD, [33] generalized anxiety disorder (GAD) [34], ADHD, [35] as well as neurological disorders like epilepsy [36, 37] and headache. [38, 39] Interestingly, acute TNS at a frequency of 120 Hz has been demonstrated to induce sleepiness and sedative effects in healthy adults while 2.5 Hz stimulation frequency did not. [40] In addition to work by Tyler and colleagues, [41] the observations made by Piquet and colleagues (2011) [40] suggest that TNS may provide some benefit for some sleep disturbances and insomnia.

Based on their initial findings, Tyler and colleagues developed Neuros with the hypothesis that repeated daily dampening of psychophysiological and biochemical arousal might improve mood if used nightly before bed, to increase the quality, duration, and/or efficiency of sleep. [41] Supportive of this hypothesis is a body of evidence that suggests that insomnia is a "waking" disorder (hyper-arousal) of RAS networks rather than a sleep disorder *per se*. [2, 42] Accordingly, a method to decrease neurobehavioral and psychophysiological arousal by stimulating trigeminal LC/RAS networks prior to sleep onset for repetitive nights can enhance the restorative qualities of sleep on mood and mental health.

2.3 Background of CES Technology

Cranial electrotherapy stimulation (CES) is the broad classification of devices that use transcutaneous electrical energy to stimulate cranial nerves. In the United States, a CES device is currently defined as a device that applies electrical current to the patient's head to treat insomnia, depression, or anxiety. The first of these devices were developed and studied in the Soviet Union in the 1950s and brought into the United States for research in the early 1960s using the name "electrosleep" to describe both the device and the therapy. [43]

After a modest number of clinical studies, the United States Food and Drug Administration (USFDA) initially classified CES devices as Class III requiring extra controls in 1979. [44] A number of devices have been introduced into the market, of which the Alpha-Stim® (Electromedical Products, Inc.) and Fisher Wallace Stimulator (Fisher Wallace Laboratories, LLC) are the two major manufacturers with medical indications for treatment of anxiety, insomnia, depression, and pain relief. These devices are also CE marked as medical devices.

Recently, the USFDA has recommended to downgrade the classification of these devices to Class II and has stated for the record that "the totality of the results of these studies (clinical evidence for CES) do provide information on the general effectiveness of CES usage for insomnia and/or anxiety." [44]

2.4 Manufacturer Data

As part of the initial product development, Tyler and colleagues published 3 pre-print open source papers of their work to clinically examine the effects of the device on safety and performance. The protocols, institutional review board (IRB) approvals, and data are all on file at Cerevast.

2.4.1 Paneri et al. 2016 [45]

This work examined the adverse effects (AEs) of CES using an early production version of the Thync Edition One. Subjects were 100 (37 females, 63 males) healthy volunteers, all > 18 years of age, with no significant health issues, not being actively treated for neuropsychiatric disorders, > 36 months since neuropsychiatric treatment, no history of head trauma, and no implants. Chronic headache or migraine sufferers were excluded. All of the procedures were embodied in a clinical study protocol approved by a local IRB.

Subjects completed an ongoing short form health survey for 2 weeks to track ongoing compliance with inclusion criteria and also completed an adverse event and adverse reaction survey following each treatment. Subjects were withdrawn in the event of atypical adverse events which were defined in the protocol and classified by occurrence as either "in session" or "between session."

Subjects were randomized to one of three groups:

1. CES using the Thync Edition One waveform (biphasic, 7-11 kHz, 5-7 mA) (n = 30),
2. tDCS using the Soterix Medical 1x1 (n = 33),
3. Sham exposure (n = 37).

All subjects remained blind to their actual treatment assignment. Sham exposure used the tDCS with initial current ramp up to 2 mA over 30 seconds and immediately back down over 30 seconds at the beginning and end of the 20 minute session. The tDCS exposure was applied with the same ramp up, with a sustained 2 mA exposure with the 30 second ramp down at the end. The CES group received 17 minutes of exposure. The CES group most closely approximates Neuros in terms of biphasic current montage (5 – 7 mA vs 4mA average in Neuros) although the frequency was significantly higher (7 – 11 kHz versus the Neuros 500 – 550 Hz). All subjects had electrodes placed in the same spot consistent with Neuros electrode application (i.e. the right supraorbital portion of the forehead and the back of the neck).

Subjects received sham or active treatments approximately 5 days per week, once per day (with a minimum of 16 hours between treatments). Treatments were administered in a relaxed open lounge area where the subjects could sit quietly and complete their personal activities (read, converse quietly, etc.). The study lasted 6 weeks and 80 subjects completed the study.

A total of 20 subjects were withdrawn from the study because of failure to meet ongoing inclusion criteria which are detailed in the table below:

Table 10.1: Summary of Compliance, Completion, and Withdrawal Rates

				Subjects that did not meet ongoing I/E criteria					
				Atypical Headache or Migraine		**Atypical Skin Condition**		**Discomfort**	
Group	**Number of Sessions**	**Total Subjects**	**Finished Trials**	**Between Session**	**Within Session**	**Between Session**	**Within Session**	**Between Session**	**Within Session**
Sham	636	37	33	2	0	2	0	0	0
tDCS	623	33	22	2	0	8	0	0	1
CES	646	30	25	3	0	1	0	0	1
Total	1,905	100	80	7	0	11	0	0	2

There were no serious adverse events with no subjects requiring medical care as a result of study participation. There were 2 subjects withdrawn from the study for within session adverse events: These were both for atypical discomfort and both cases were attributed to non-ideal electrode positions which in turn lowered the contact area and raised the current density leading to the discomfort. The remaining withdrawals occurred as a result of between session reports. The table below details the incidence rates of adverse events reported in all groups.

Table 10.2: Summary of During Stimulation Adverse Events

Group→ / Event Type↓	Sham	tDCS	CES	Notes
Mild Tingling (Within Session)	70.2% ± 1.8%	55.7% ± 2.0%	25.8% ± 1.7%	Sham > tDCS ($p < 0.01$) Sham > CES ($p < 0.01$)
Mild Burning (Within Session)	27.7% ± 1.8%	23.3% ± 1.7%	3.4% ± 0.7%	Sham > tDCS ($p < 0.01$) Sham > CES ($p < 0.01$)
Itching Sensation (Within Session)	29.5% ± 1.8%	30.9% ± 1.9%	13.5% ± 1.3%	No statistical differences
Headache (Within Session)	3.9% ± 0.8%	4.4% ± 0.8%	2.6% ± 0.6%	No statistical differences
Headache (Between Session)	2.4% ± 0.6%	1.3% ± 0.5%	1.2% ± 0.4%	No statistical differences
All others	< 5%	< 5%	< 5%	No statistical differences

There were no statistical differences in occurrence which indicated that active (either CES or tDCS) therapy produced significantly more adverse effects than sham. Specifically, the incidence of adverse events in the sham group exceeded that of the active groups for mild tingling and mild burning.

The tingling, burning, and itching sensations are all cutaneous nociceptive due to stimulation of cranial and cervical spinal nerve afferents that is related to electrode electrochemical performance and skin current flow. These events occurred at a lower rate in the CES group which indicates tolerability of the waveform and electrode configuration. The absence of significant differences in headache sensations between all groups, including sham, indicates that CES does not add any additional risk of headache nor other AE occurrences.

2.4.2 Tyler et al. 2015 [41]

This work was completed with the Thync Edition One device using 7 – 11 kHz and 5 - 7 mA and 50% duty cycle. The work was organized into 3 experiments. All subjects were healthy volunteers with no neurological or psychiatric disorders, no severe face or head trauma, no cranial or facial implants, no high blood pressure, no diabetes, nor heart disease. All of the procedures were embodied in a clinical study protocol approved by a local IRB.

2.4.2.1 Experiment 1

This experiment studied the effect of CES on facial infrared thermography with the hypothesis that CES exposure would increase facial temperature as a result of decreased sympathetic activity and a increased blood flow due to facial blood vessel dilation.

A total of 19, right-handed healthy volunteers were randomized to receive CES (n = 9) or sham (n = 10). There were no significant differences between subjects. All subjects were fitted with the CES device without start of treatment and 5 minutes of baseline infrared thermography data was collected to determine starting temperatures at the forehead, chin, cheek, and nose.

There were no significant differences between groups during the baseline period. All subjects were subsequently given 15 minutes of sham or actual CES treatment.

Both sham and CES produced significant increases in temperature at all 4 monitored facial regions. Compared to sham, the temperature increases were greater for TEN across the times measured for the forehead, chin, and cheek. The results tabulated below show statistics comparing sham to treatment temperature changes. There were significant differences between the groups for temperature increase at the forehead, chin, and cheek. The temperature increase at the nose was higher for the CES group, although this difference was not significant.

Table 10.3: Summary of Temperature Increase

Location→	Forehead	Chin	Cheek	Nose
ANOVA [a]	(4,68) 4.73	(4,64) 4.61	(4,68) 8.26	(4,68) 2.18
p-value	p = 0.046	p = 0.012	p = 0.001	p = 0.123

2.4.2.2 Experiment 2

Experiment 2 was designed to test the effect of CES on the Profile of Moods State (POMS) survey.[46] There were 45 healthy volunteer subjects randomized to receive CES (n = 25) or sham (n = 20). Subjects were given a 5 minute baseline with no treatment in a testing booth, followed by 15 minutes of CES or sham treatment, then a 10 minute rest period before answering the POMS questionnaire.

There were significant differences in the Tension-Anxiety (POMS-TA) subscale, but none of the other groups as shown in the table below.

Table 10.4: Summary of Post-Treatment POMS

POMS Sub-Scale→ Group↓	Tension-Anxiety	Anger-Hostility	Depression-Dejection	Fatigue-Inertia	Vigor-Activity
CES	1.73±0.52	-	-	-	-
Sham	2.10±0.64	-	-	-	-
ANOVA[a]	(1,44) 4.51	(1,44) 1.96	(1,44) 3.11	(1,44) 0.206	(1,44) 0.19
p-value	0.04	0.169	0.085	0.652	0.613

These data show that CES significantly reduced self-reported tension and anxiety compared to sham in the absence of other environmental stimuli. However, it should be noted that POMS was not administered to subjects before therapy, thus it cannot be confirmed that there were no known differences in this regard between the groups *a priori*.

2.4.2.3 Experiment 3

Experiment 3 was designed to test different measures of stress and the effects of CES concurrent with an acute stress induction. A total of 20 healthy volunteers, all males were randomized evenly (n = 10) between CES and sham. Only males were enrolled to control for confounding factors due to hormonal variance across menstrual cycles on stress biochemistry. All treatments took place between 13:00 and 16:00 to limit circadian variation factors. The CES parameters were the same as the preceding 2 experiments.

All subjects were fitted with a heart rate variability (HRV) monitor and galvanic skin conductance (GSC) sensors.

The steps within the experiment, for all subjects, are shown in the figure below.

Figure 10.1: Experiment 3 Schedule of Events

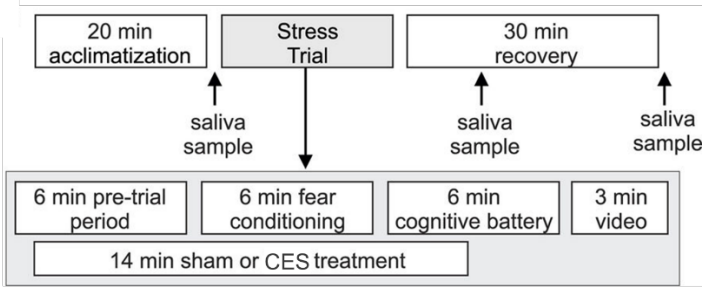


Image from Tyler et al. 2015

Subjects were acclimated for 20 minutes before drawing of the baseline saliva sample. The stress trial started 30 seconds before either sham or actual CES treatment, which commenced for 14 minutes. In the first 6 minutes, the pre-trial period allowed for the effects of CES to build up, followed by a 6 minute fear-conditioning paradigm, followed by the timed cognitive tests (3 tests, each 2 minutes),

[a] ANOVA statistic expressed as (between group degrees of freedom (df), within group df) F-test score.

followed by a 3 minute video of a nature scene. During the recovery period two more saliva samples were drawn at 10 minutes and 30 minutes post treatment.

Results are shown below in the following two tables.

Table 10.5: Experiment 3 Physiological Results

Category	Measure	Time Point	CES	Sham	p-value[a]
HRV Tests	HR (bpm)	Stress Trial	69.18 ± 7.34	67.01 ± 8.47	0.55
	R-R Int. (msec)		885.21 ±94.34	922.07 ± 124.88	0.466
	SDNN (msec)		78.48 ± 16.60	105.45 ± 28.91	0.02
	LF Power ($msec^2$)		1567.40 ± 827.24	2946.80 ± 1352.12	0.01
	HF Power ($msec^2$)		1381.50 ± 804.42	2367.70 ± 1534.22	0.09
	LF/HF Ratio		1.20 ± 0.50	1.42 ± 0.43	0.30
GSC Tests	ΔGSC_{fear}	Stress Trial	0.37 ± 0.14	0.54 ± 0.11	0.007
	ΔGSC_{shock}		0.09 ± 0.06	0.19 ± 0.04	0.001
Saliva Tests	α-amylase (U/mL)	Baseline	85.57 ± 41.03	102.45 ± 39.60	0.39
		10 min post-tx	68.93 ± 27.26	103.13 ± 46.43	0.07
		30 min post-tx	68.62 ± 28.93	110.30 ± 35.11	0.01
	Cortisol (μg/dL)	Baseline	0.24 ± 0.09	0.26 ± 0.13	0.75
		10 min post-tx	0.19 ± 0.11	0.23 ± 0.15	0.51
		30 min post-tx	0.16 ± 0.06	0.18 ± 0.09	0.67

Notes regarding HRV measures:
The SDNN Standard Deviation of the normal-to-normal (NN) interval is a measure of overall variability.

R-R Interval is the time between repeating sinus rhythm from the "R" peak to the next successive "R" peak.

LF Power (0.04 – 0.15 Hz) band is thought to be a marker of sympathetic and vagal influence

HF Power (0.15 – 0.4 Hz) band reflects parasympathetic or vagal activity and is frequently called the respiratory band because it corresponds to the HR variations related to the respiratory cycle known as respiratory sinus arrhythmia.

Among the HRV tests, only the SDNN and LF power tests showed significance ($p < 0.05$).

Table 10.6: Experiment 3 Cognitive Tests

Test	Measure	CES			Sham			Test/Remarks[b]
		n	$\bar{x}$	sd	n	$\bar{x}$	sd	
Flanker	% Correct	10	100	0	10	99	2.11	F(1, 19) = 2.25, p = 0.15
	Congruent RT	10	612.37	249.27	10	570.43	190.43	F(1, 19) = 0.18, p = 0.68
	Incongruent RT	10	585.92	170.93	10	618.00	182.06	F(1, 19) = 0.17, p = 0.69
Stroop	% Correct	8	98.75	3.54	6	97.82	2.79	F(1, 13) = 0.28, p = 0.61
	Congruent RT	8	973.41	305.72	6	931.88	132.58	F(1, 13) = 0.10, p = 0.76
	Incongruent RT	8	1247.82	289.62	6	1102.69	170.31	F(1, 13) = 1.18, p = 0.30
N-Back	% Correct	8	78.75	10.69	9	83.75	13.42	F(1, 16) = 0.71, p = 0.41
	Average RT	8	705.80	103.74	9	713.25	133.01	F(1, 16) = 0.02, p = 0.90
	Combined Time	8	921.80	245.53	9	890.29	291.78	F(1, 16) = 0.06, p = 0.81

None of the cognitive tests showed significance.

[a] All t-tests
[b] One-Sided multiple regression, analysis of variation, (MRANOVA) test, shown as "F([degrees of freedom of treatment], [degrees of freedom of total error]) = [F-statistic value], p = [p-value]"

2.4.3 Boasso et al. 2016 [1]

This work directly applies to Neuros as it used the same device, with the similar waveform parameters (LF-only from Experiment #3). The work was organized into 3 experiments.

2.4.3.1 Experiment 1

This was designed to examine the effects of CES using Thync Edition One (directly equivalent to Neuros except for waveform frequency in some experiments) before bedtime. This was designed as a single group, prospective study in which each subject was observed for a week for baseline data (no device intervention) followed by a week of CES therapy with the device before bedtime. Of the 43 subjects enrolled, 38 completed the two-week regimen (17 females, 21 males. Mean age 29.68 ± 10.88 years. Age range 20 – 62 years).

During both weeks, the subjects self-reported their mood each morning with the Positive and Negative Affectivity Scale (PANAS) [47] and ratings of awakening drowsiness and refreshment. At the end of each week the subjects completed the self-reported Depression, Anxiety and Stress Scale (DASS). The DASS is a reliable and clinically validated measure which is measured on a "0" to "3" point severity or frequency scale indexed to the previous week across 42 items. [48, 49] The 42 items are sub-divided into three categories, 14 items each, of depression, anxiety, and stress.

The interventional week of the treatment included 20 minutes of treatment with the device, once per day, for 7 consecutive days. The treatment was administered within 30 minutes before going to bed and consisted of a pulse modulated (3 – 11 kHz) biphasic electrical current producing average amplitudes of 5 – 7 mA applied for 20 minutes.

All measures showed significant improvement except the DASS Depression Score. The results are shown in the table below.

Table 10.7: Results of Boasso et al. 2016 Experiment 1

Measure	Scale	Baseline Week $\bar{x} \pm sd$	CES Week $\bar{x} \pm sd$	Test / Remarks[a]
Drowsiness	Drowsiness & Refreshment scale	3.97 (1.63)	2.89 (1.28)	t(36) = -4.859, p < 0.001
Refreshment		4.88 (1.89)	5.79 (1.74)	t(36) = 4.908, p < 0.001
Negative Affect	PANAS [47]	2.23 (0.83)	2.42 (0.85)	t(35) = -3.147, p = 0.003
Positive Affect		1.31 (0.27)	1.17 (0.17)	t(35) = 3.427, p = 0.002
Depression	DASS [48]	4.07 (5.35)	3.43 (5.65)	t(29) = -0.733, p = 0.469
Anxiety		3.10 (2.71)	2.17 (3.12)	t(29) = -2.177, p = 0.038
Stress		10.00 (6.96)	5.87 (5.31)	t(30) = -3.982, p < 0.001

The results of Experiment 1 provide clear evidence of the effectiveness of CES over controls for the alleviation of anxiety, as well as morning drowsiness & refreshment and affect. There were no reported adverse events in Experiment 1.

2.4.3.2 Experiment 2

Experiment 2 was designed as a follow-on study to Experiment 1 and was a randomized, controlled, double-blinded study of actual CES vs. sham CES. Each subject started with a week of baseline reporting followed by a week of actual or sham CES. There were 42 subjects enrolled, of which 26 (14 actual CES, 12 sham CES) completed the two-week study (62.1% females, 37.9% males. Mean age 27.66 ± 9.99 years. Age range 19 – 59 years).

During both weeks, each morning the subjects completed the PANAS, rated their waking drowsiness and refreshment, reported their number of awakenings, and rated their overall sleep quality. At the end of both weeks, each subject completed the DASS. Additionally, all subjects wore a clinically validated actigraph to track sleep and wake cycles (Phillips Actiwatch) [50, 51] and a heart rate monitor to track heart rate variability and ECG data (Polar H7).

The interventional week of the treatment included 20 minutes of treatment with the device (either sham or actual), once per day, for 7 consecutive days. The treatment was administered within 30

[a] Two-sided t-test, shown as "t(degrees of freedom) = [t-value], p=[p-value]"

minutes of going to bed and consisted of a pulse modulated (3 – 11 kHz) biphasic electrical current producing average amplitudes of 5 – 7 mA applied for 20 minutes.

All measures showed significant improvement except the DASS Depression Score PANAS Affect scores. The results are shown in the two tables below.

Table 10.8: Experiment 2 Baseline to Treatment Comparisons

Measure	Scale	Sham CES Group		p-value	Actual CES Group		Test / Remarks[a]
		Baseline Week $\bar{x} \pm sd$	Sham Week $\bar{x} \pm sd$		Baseline Week $\bar{x} \pm sd$	CES Week $\bar{x} \pm sd$	
Drowsiness	Drowsiness & Refresh. scale	4.35 (1.30)	3.91 (1.15)	p> 0.200	4.69 (1.59)	3.80 (1.63)	t(17) = 4.859, p < 0.001
Refreshment		3.77 (1.06)	3.89 (0.87)	p> 0.200	3.89 (0.76)	4.41 (0.78)	t(17) = 4.908, p < 0.001
Negative Affect	PANAS [47]	2.17 (0.58)	2.30 (0.72)	p> 0.200	2.04 (0.52)	2.26 (0.64)	t(17) = -0.237, p = 0.815
Positive Affect		1.40 (0.40)	1.41 (0.51)	p> 0.200	1.28 (0.28)	1.27 (0.034)	t(17) = 4.859, p < 0.001
Sleep Quality	Self-reported	4.58 (0.70)	4.36 (0.96)	p> 0.145	4.51 (0.90)	5.01 (0.89)	t(17) = 2.155, p = 0.046
# Awakenings		1.41 (1.23)	1.20 (1.40)	p> 0.145	1.19 (0.84)	0.75 (0.62)	t(17) = -3.351, p = 0.003
% ΔSleep Time	Actigraph (minutes)	-8.5 (9.61)			+14.1 (6.0)		t(14) = 2.255, p = 0.041
% ΔTime Awake		-4.5 (2.51)			-11.2 (3.12)		t(14) = -2.329, p = 0.035

Table 10.9: Experiment 2 Group Comparisons

Measure	Scale	Sham CES Group[b]	Actual CES Group[b]	Test / Remarks[c]
% Δ Depression	DASS [48]	-33%	-30%	F(1,25) = 0.091, p = 0.766
% Δ Anxiety		+19%	-41%	F(1,25) = 4.392, p = 0.047
% Δ Stress		-12%	-44%	F(1,25) = 4.907, p = 0.036

The results of Experiment 2 provide clear evidence of the effectiveness of CES over controls for the alleviation of anxiety and insomnia. There were no reported adverse events in Experiment 2.

2.4.3.3 Experiment 3

This was a randomized, controlled, double-blinded, crossover design with 25 total subjects enrolled to evaluate the effectiveness of a lower pulse frequency (CES_{LF}) at 500 – 750 Hz and current < 5 mA. The hypothesis to be tested was whether CES_{LF} would have a better effect at improving sleep and reducing stress than CES_{HF} (3 – 11 kHz, 5-7 mA) which had been evaluated in Experiments 1 and 2 discussed above.

The subjects were randomized into an active treatment group receiving active CES_{HF} or sham CES during the first 7 days each night before bedtime. The subjects were then crossed over to receive 7 days of CES_{LF} or a control group which concurrently received CES_{HF}.

The subjects wore the same actigraph described in Experiment 2 each night to track key sleeping parameters and manually entered sleep log data in the morning upon awakening. The design of experiment 3 is shown in the figure below.

Figure 10.2: Experiment 3 Design

[a] Two-sided t-test, shown as "t(degrees of freedom) = [t-value], p=[p-value]"
[b] These values approximated from the graphs in the printed version
[c] One-Sided multiple regression, analysis of variation, (MRANOVA) test, shown as "F([degrees of freedom of treatment], [degrees of freedom of total error]) = [F-statistic value], p = [p-value]"

The results of week 1 (CES_{HF} vs. Sham) were consistent with previously observed results and are summarized in the table below.

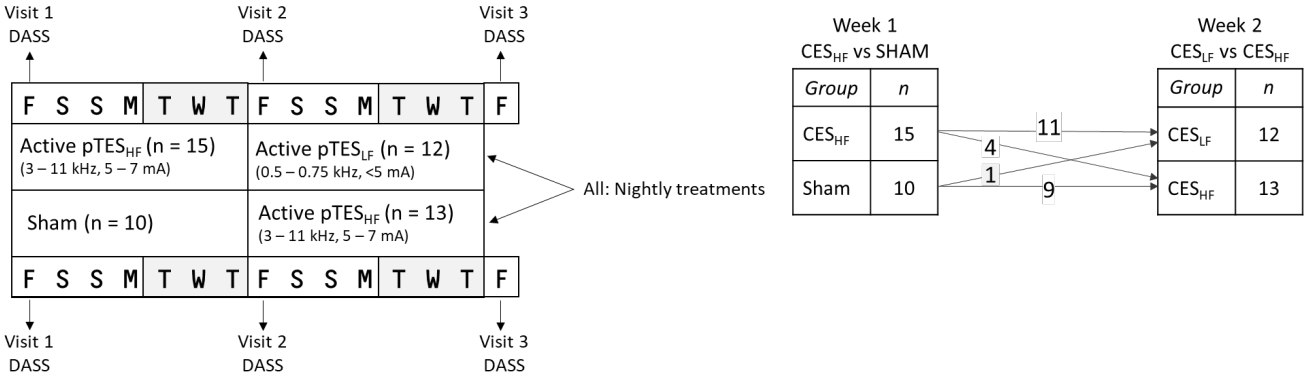


Table 10.10: Experiment 3 Group Comparisons (Week 1)

Measure	**Scale / Measure**	**CES_{HF} Group**	**Sham Group**	**Test / Remarks[a]**
Number of Wakeups	Actigraphy	22.4±1.88	24.8±1.88	t(19) = 3.781, p = 0.001
Wake after Sleep Onset (minutes)		42.7±3.46	48.5±4.68	t(19) = -2.381, p = 0.028
% Time Awake		10.25±2.10	11.3±2.62	t(19) = -2.241, p = 0.037
Δ Mean Depression	DASS [48]	-	-	t(20) = -1.418, p = 0.172
Δ Mean Anxiety		-	-	t(20) = -3.406, p = 0.003
Δ Mean Stress		-	-	t(20) = 0.670, p = 0.510

The results of week 2 (CES_{LF} vs. CES_{HF}) were consistent with previously observed results and are summarized in the table below.

Table 10.11: Experiment 3 Group Comparisons (Week 2)

Measure	**Scale / Measure**	**CES_{LF} Group[b]**	**CES_{HF} Group[b]**	**Test / Remarks[c]**
Number of Wakeups	Actigraphy	21.9±3.50	24.2 ±3.92	t(9) = 2.796, p = 0.021
Wake after Sleep Onset (minutes)		43.5 ±6.41	52.9 ±8.85	t(9) = 2.808, p = 0.020
% Time Awake		10.7 ±2.02	12.3 ±1.67	t(9) = 3.006, p = 0.015
Δ Mean Depression	DASS [48]	-	-	t(8) = -0.769, p = 0.467
Δ Mean Anxiety		-5.31 ±1.30	-2.91 ±0.71	t(8) = -2.382, p = 0.044
Δ Mean Stress		-	-	t(8) = 0.928, p = 0.381

The results show a clear superiority of CES_{HF} over sham, and CES_{LF} over CES_{HF}. These results speak directly to the effectiveness of CES in general and CES_{LF} specifically (essentially the Neuros waveform and current), for the alleviation of anxiety and insomnia. There were no reported adverse effects from Experiment 3.

2.4.4 Summary of Results for Data Held by the Manufacturer

The collective results of Paneri et al. 2016, Tyler et al. 2015, and Boasso et al. 2016 provide adequate evidence for the safety and performance profile of Neuros pTES.

[a] t-tests presented as "t([degrees of freedom]) = [t-value], p = [p-value]"
[b] These values approximated from the graphs in the printed version
[c] ibid

2.5 Clinical Data Retrieved from Literature

2.5.1 Scope of Literature Search

The scope of the literature search was to seek out clinical, safety, and performance data related to the treatment of sleep and anxiety disorders with non-invasive electrical stimulation applied to the nerves that innervate the skin of the head and neck.

There are many other forms of non-invasive application of electrical stimulation to the head for inducing a neurological effect. These methods include, but are not limited to, transcranial direct current stimulation (tDCS), transcranial alternating current stimulation (tACS), electroconvulsive therapy (ECT), electroanesthesia (EA), and many others. Based on the current dose and application of Neuros electrodes to the patient, it was decided to limit the scope of search to those studies which employed cranial electrical stimulation (CES) and/or electrosleep. Some background is in order to explain why the search was contained to these classifications.

The emergence of the term electrosleep was associated with clinical studies published from 1902 to 1914 [43]. Much of the following clinical work took place in Russia until the 1960s when researchers in Europe and the United States took notice and conducted their own research. In 1966, the first symposium on electrosleep and electroanesthesia was conducted in Graz, Austria and it was when researchers agreed that electrosleep does not actually induce sleep, rather these devices reduce anxiety and impart a calming effect. Soon afterwards, the term cranial electrostimulation therapy (CET) came into use, although the term electrosleep persisted in United States research publications through much of the 1970s. In 1978, the United States FDA classified electrosleep devices as Class III for the treatment of anxiety, insomnia, and depression. As part of this classification, the term cranial electrical stimulation (CES) was adopted by the FDA and remains broadly in use today by researchers, manufacturers, and regulators.

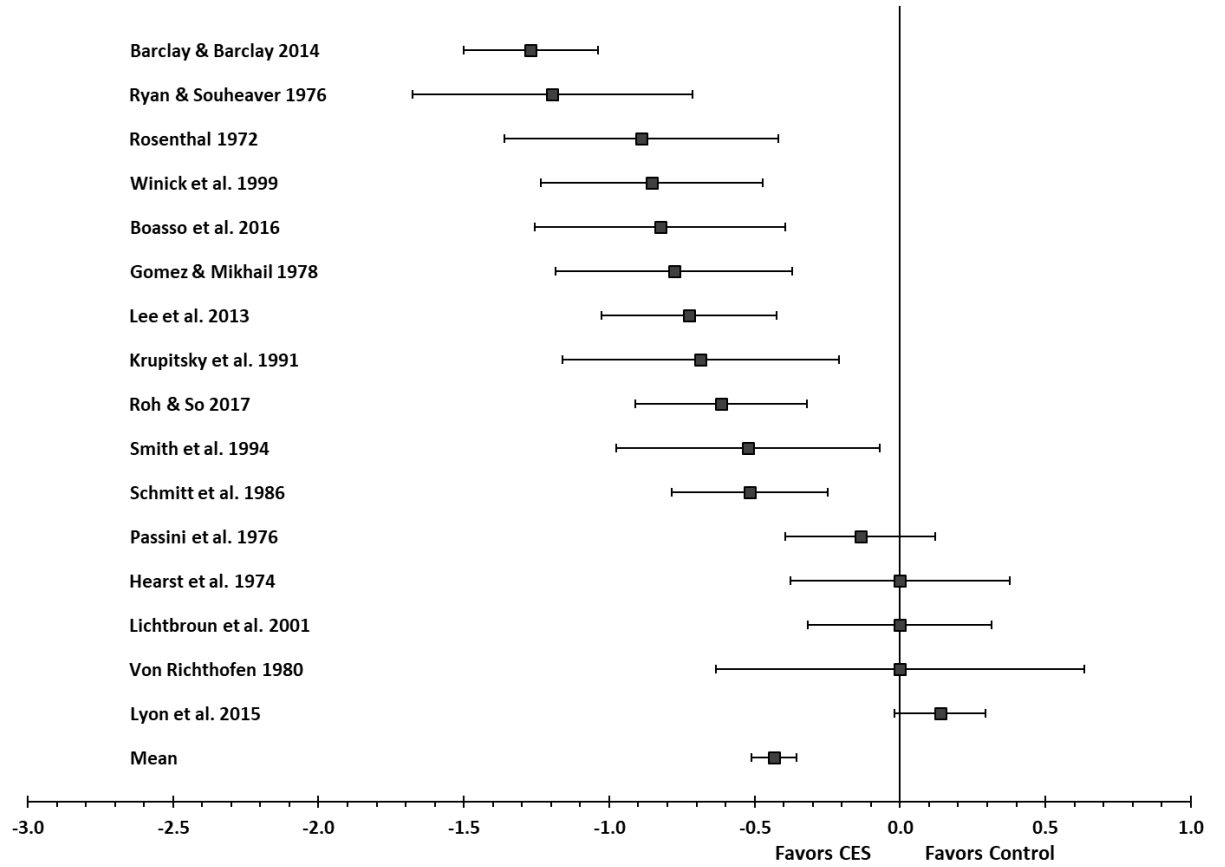
Based on the electrical waveform montage (peak current levels being the most critical, but also frequency, and duty cycle) and electrode placement, Neuros most closely resembles the CES devices and their electrosleep predecessors.[a] Accordingly, as part of the clinical evaluation for the introduction of this product, we undertook a comprehensive review of hundreds of published clinical studies using CES and their conclusions. A strict set of exclusion criteria were applied to the body of research and a meta-analysis applied. A summary of meta-analysis for CES devices application for treatment of Anxiety and Insomnia as follows:

2.5.2 Anxiety Meta-Analysis

2.5.2.1 The effect size plot for CES treatment of anxiety is shown below. Each of the studies listed was a randomized, controlled, blinded (at least patient blinding) study to evaluate CES for the treatment of anxiety vs a sham control. Each of the horizontal bars represents the effect size, noted by the square, and 95% confidence interval of effect size shown as the bar on either side.

Figure 12.1: Effect Size Plot of CES for Treatment of Anxiety

[a] Based on the current doses and electrodplacements shown in Guleyupoglu et al. 2014 **43**



2.5.2.2 By convention, a negative treatment effect tends to favor the experimental therapy; CES. This is because most relevant measures of anxiety use a score which increases with anxiety, thus a therapy which reduces anxiety more so than the control therapy will have a negative effect size. Conversely, a positive effect size favors the control therapy. The studies are ordered by rank of highest to lowest effect size for ease of visual integration of the data. Each study is depicted as a bar centered about the computed effect size bounded by the 95% confidence interval. At the bottom, the mean effect size, which is a mean of all of the studies listed above it, is shown with corresponding 95% confidence interval. For example, Barclay and Barclay 2014 has an effect size of -1.2699, with 95% CI bounds at -1.7223 (Upper Limit) and -0.8175 (LL).

2.5.2.3 The mean effect size for all studies is -0.4339 (-0.5858, -0.2820). In terms comparison of relative effect, Cohen, and later, Sawilowsky, provided rules of thumb to gage effect size. (Cohen, 1988), (Sawilowsky, 2009)

Table 12.3: Descriptors of Effect Size

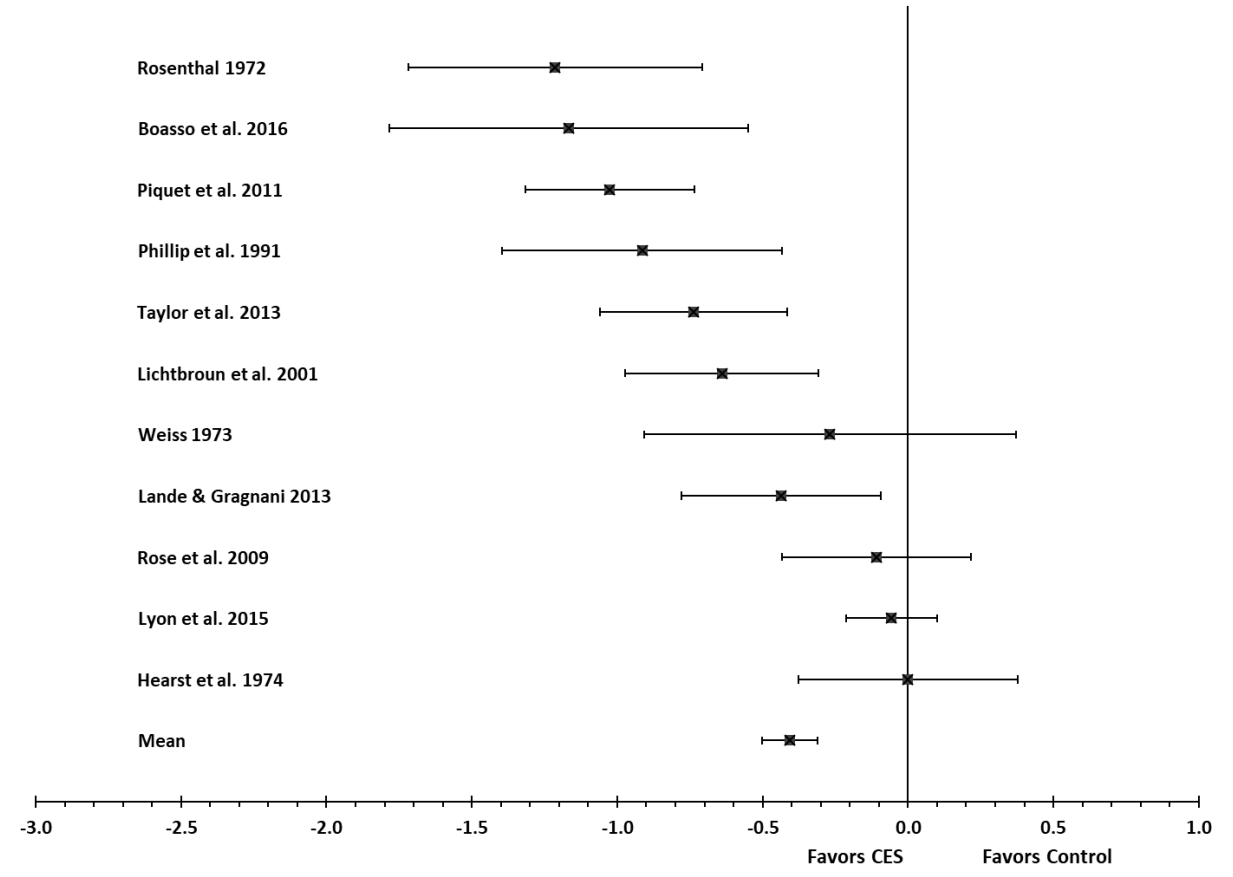
Effect Size	Descriptor	Source
0.01	Very Small	Sawilowsky 2009
0.20	Small	Cohen 1988
0.50	Medium	Cohen 1988
0.80	Large	Cohen 1988
1.20	Very Large	Sawilowsky 2009
2.00	Huge	Sawilowsky 2009

It can inferred that the observed treatment effect for the indication of anxiety is slightly below “medium” in favor of CES over controls.

2.5.3 Insomnia Meta-Analysis

2.5.3.1 Figure 12.2 is the effect size plot for CES treatment of insomnia. As with anxiety above, each of the studies listed was a randomized, controlled, blinded (at least patient blinding) study to evaluate CES for the treatment of insomnia vs. a sham control.

Figure 12.2: Effect Size Plot of CES for Treatment of Insomnia



2.5.3.2 The overall observed treatment effect for all of the studies is -0.4061 (-0.5934, -0.2188) which can be judged as just below "medium" in favor of CES treatment over controls.

CONCLUSION

When used as directed, Neuros Pulsed Transdermal Electrical Stimulation (pTES) and CES are proven to provide safe and effective treatment for anxiety and insomnia. Neuros pTES offers a non-pharmaceutical alternative for relief from these neurological ailments.

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